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Multi-Year Assessment of Airborne Metals in Fallon, Nevada, using Leaf-Surface Chemistry

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1. Introduction

1.1 Fallon, Nevada

Spatial patterns of airborne metals are described from leaf-surface chemistry of trees in Fallon, Nevada (Fig. 1a), where a cluster of childhood leukemia began in 1997. Officially, 16 cases of childhood leukemia were diagnosed from 1997 to 2002 inclusive (Expert Panel, 2004), and one additional case was reported in December 2004 (Nevada State Health Division, 2004). Although the cluster is thought to have abated (Reno Gazette-Journal, 11 October 2008), at least one additional case of childhood leukemia has occurred in Fallon since 2004 (Lahontan Valley News, 15 October 2010). Given Fallon's pediatric population of about 2500 children up to 19 years in age (U.S. Census, 2000) and a national expected rate of childhood leukemia of 4.1 cases per 100,000 children up to 19 years in age per year (U.S. NCI, 2003), the expected rate of childhood leukemia for Fallon should be only one case every ten years.

This cluster, deemed "one of the most unique ever reported" (Steinmaus et al., 2004; Steinmaus et al., 2005), prompted multiple investigations to determine if an environmental cause might have been responsible. Research focused on drinking water (Moore et al. 2002; Shaw et al., 2005; Walker & Fosbury 2009; Walker et al., 2006), jet fuel (U.S. ATSDR, 2002), pesticides (Rubin et al., 2007; U.S. CDC, 2003), surface water (U.S. ATSDR, 2003a), outdoor air (U.S. ATSDR, 2003b), surface soil and indoor dust (U.S. ATSDR, 2003c), potential lingering effects of underground nuclear bomb testing in the area (Seiler, 2004), and groundwater (Seiler et al., 2005). A non-environmental hypothesis–population mixing–was also considered (Kinlen & Doll, 2004; Wakeford, 2004]. Few definitive conclusions emerged from these studies, prompting an interpretation that Fallon had been given a "clean bill of health" by the US Agency for Toxic Substances and Disease Registry and the US Centers for Disease Control (Lahontan Valley News, 8 August 2007). However, this interpretation is questionable (Pleil et al., in press), and the need to monitor the environment of Fallon continues to exist.

To monitor the environment of Fallon, we have employed multiple techniques, all of which have shown notable patterns of airborne tungsten and cobalt. Elevated tungsten and cobalt was identified in airborne particulates of Fallon relative to comparison towns (Sheppard et al., 2006a) and in lichens within Fallon compared to outlying desert areas (Sheppard et al.,

2007b). Tungsten and cobalt maxima were found in surface dust (fallout from air) near the center of Fallon, just north and west of the crossroads of the main highways (Sheppard et al., 2007a). Dendrochemistry showed that tungsten began increasing in Fallon tree rings by the mid-1990s, coinciding roughly with the onset of the cluster of childhood leukemia (Sheppard et al., 2007c). From direct microscopy analysis of airborne tungsten particles in Fallon, they are anthropogenic in origin, not natural (Sheppard et al., 2007d).

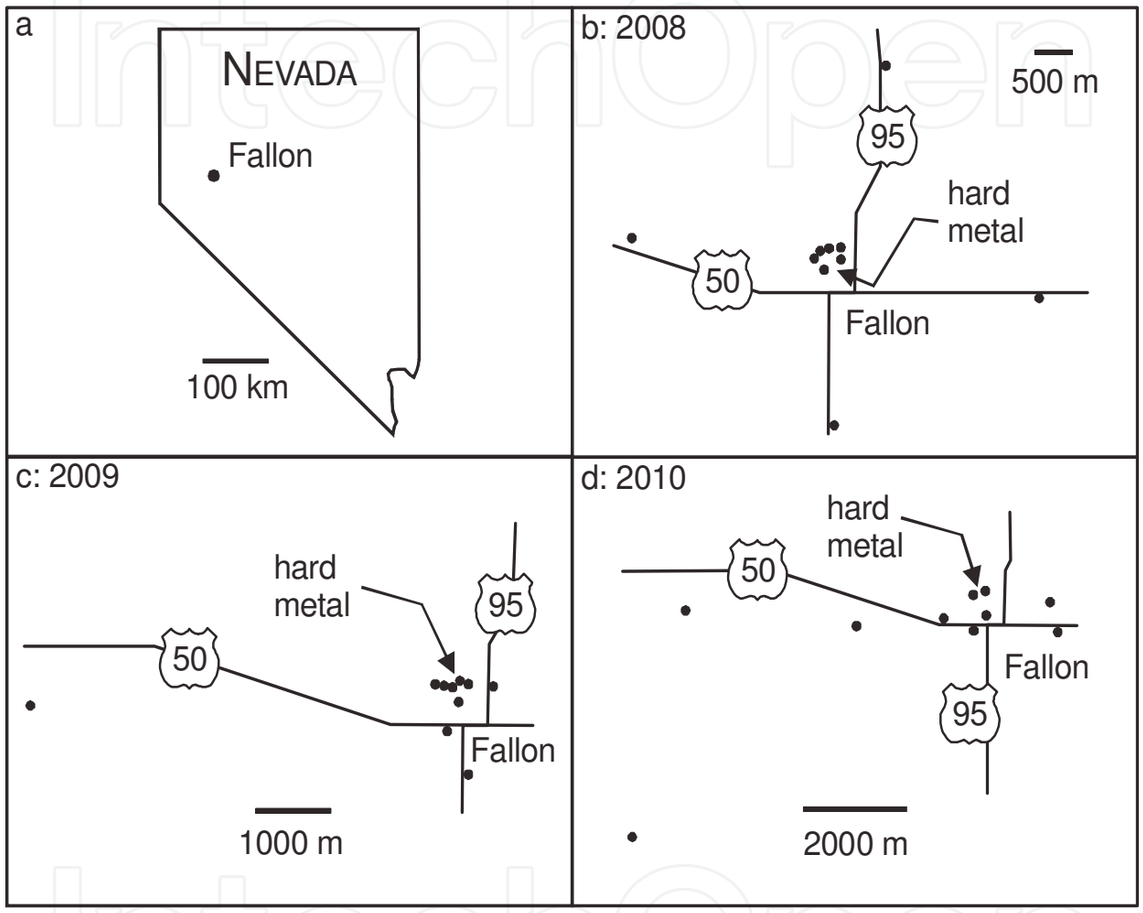

Fig. 1. Maps of Nevada, showing Fallon (a) and of Fallon, showing trees from which leaves were collected each year from 2008 through 2010 (b-d). An industrial facility performing hard-metal metalurgy is located just northwest of the main intersection of Highways 95 and 50.

1.2 Leaf-surface chemistry to assess air quality

An environmental monitoring technique that is applicable for assessing air quality is leaf-surface chemistry, the measurement and interpretation of element concentrations in particulates that accumulate on surfaces of leaves of trees and other plants. Leaf-surface chemistry indicates atmospheric chemistry (Wittig, 1993), including airborne metals (Rautio et al., 1998). Leaves are easy to collect (Aksoy et al., 1999), so spatial and temporal arrays of samples can be obtained quickly (Loppi et al., 1997). Leaf-surface particulates reflect the chemical composition of recent accumulations, on the order of weeks to months or perhaps an entire growing season depending on the occurrence of precipitation (Alfani et al., 1996b).

By collecting leaves across a region, differing accumulations of airborne metals can be mapped, thereby pinpointing source areas (Aboal et al., 2004). Paired studies of leaf-surface accumulations with ground-surface dust and/or airborne particulates can be particularly fruitful for confirming airborne chemistry and identifying spatial patterns of metals (Bargagli 1993; Čeburnis and Steinnes 2000). Because of these advantages, many case studies exist worldwide using leaf-surface chemistry to quantify atmospheric loading of heavy metals and/or identify their spatial patterns (e.g., Aksoy and Öztürk, 1997; Aksoy et al., 1999; Alfani et al., 1996a; Dasch, 1987; Gupta et al., 2004; Rossini Oliva and Mingorance, 2006; Salve et al., 2006; Ward, 1977;). More specifically, leaf-surface chemistry was done in Fallon in 2007, and it showed elevated peaks of airborne tungsten and cobalt just northwest of the center of town (Sheppard et al., 2009a).

1.3 Objective

For years now, we have been collecting various biological samples in and around Fallon for the purpose of assessing air quality there. Total suspended (airborne) particulate samples were collected in March and November 2004. Lichens and surface dust were collected in March 2005. Tree leaves were first collected in October 2007. Tree-ring samples have been collected multiple times since 2002. As time progresses, this time series of observations of air quality of Fallon becomes more and more precious by virtue of the ability to discern change in air quality through time. Therefore, it is important that regular monitoring of air quality of Fallon be maintained. A relevant Nevada state agency charged with environmental monitoring and protection has stated no plans for doing investigations in Fallon (Lahontan Valley News, 3 December 2005), but we are carrying on with environmental monitoring in Fallon.

Weighing advantages and disadvantages of various environmental monitoring techniques (Sheppard et al., 2009b), we have chosen leaf surface chemistry as the technique with which to continue monitoring Fallon air quality. Accordingly, leaves from trees of Fallon were collected in October of 2008, 2009, and 2010. The objective of this chapter is to describe the surface chemistry of these leaves to update spatial patterns of airborne tungsten and cobalt in Fallon.

2. Methods

2.1 Site description

Fallon is a small, rural, farming community (Greater Fallon Area Chamber of Commerce, 2008) located in west-central Nevada (Fig. 1a). Its climate is cool to mild and dry, with a mean annual temperature and precipitation of 10.7° C and 127 mm, respectively, as typified from meteorological data from Fallon (monthly data from 1931 to 2010 obtained on-line from the National Climatic Data Center, NOAA 2010). Along with service industries and small businesses, Fallon has a facility that does hard-metal metallurgy, which includes tungsten carbide and cobalt (Harris and Humphreys, 1983). The hard-metal facility has been considered a candidate source of tungsten within Fallon generally (Reno Gazette-Journal, 5 February 2003) and more specifically of elevated tungsten and cobalt in total suspended particulates and in surface dust of Fallon (Sheppard et al., 2006a, 2007a).

2.2 Leaf collection

Tree leaves were collected in mid- to late-October of the years 2008, 2009, and 2010. All trees sampled were deciduous species, so results of this study reflect accumulations of airborne particulates onto leaf surfaces during just the growing season of each year. During the leaf

season (May–October) of all four years 2007–2010, measurable rainfall was recorded in varying amounts at the nearby Fallon Naval Air Station (Fig. 2; daily data obtained on-line from the National Climatic Data Center, NOAA 2010).

All trees sampled were broadleaf species, in part because conifer species are not common in Fallon but also because broad leaves provide ample surface for accumulating airborne particulates. Tree species was not held constant during collecting because no single species predominates throughout all parts of Fallon. The urban forest of Fallon contains many kinds of trees, and elm (*Ulmus*), mulberry (*Morus*), cottonwood (*Populus*), and ash (*Fraxinus*) were the most common tree types sampled. Accumulation of aerosols onto leaves can be affected by leaf characteristics such as roughness, pubescence, moisture, and stickiness (Wedding et al., 1977), but these characteristics of leaves do not vary appreciably across the tree species sampled in this study.

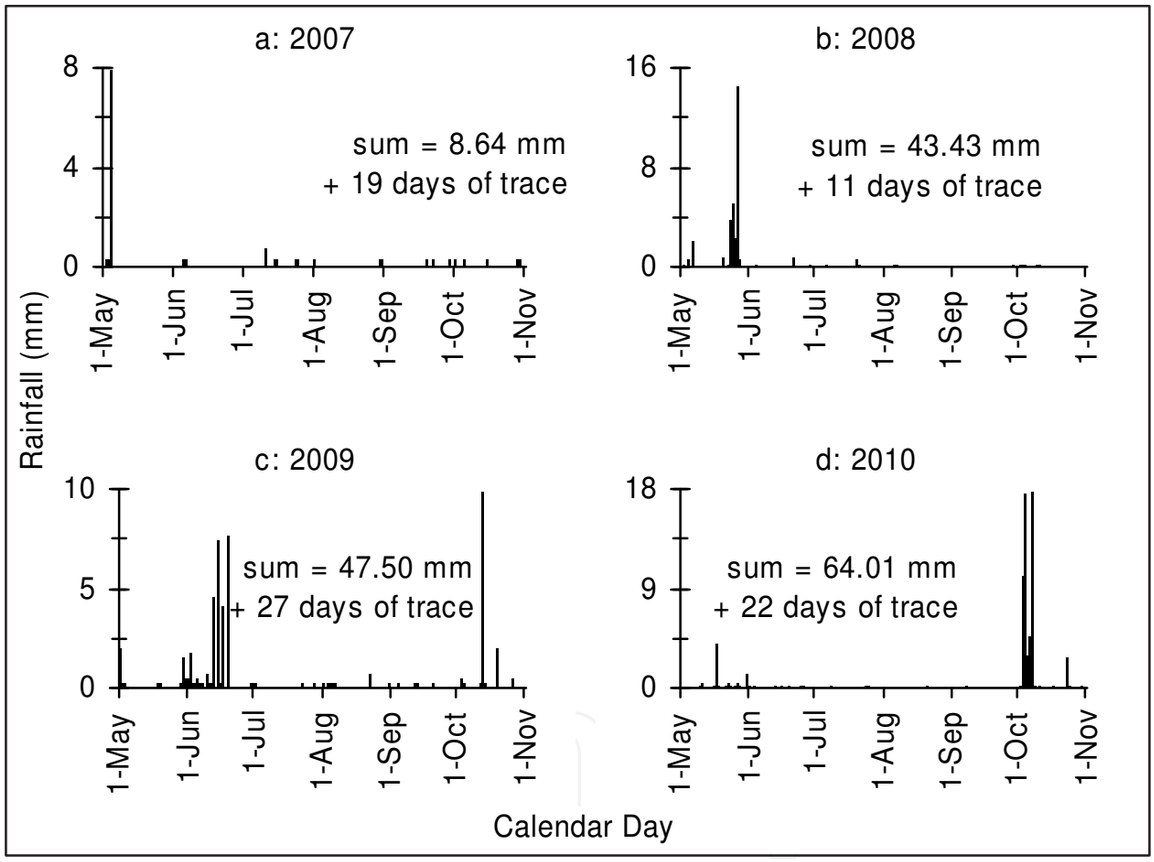

Fig. 2. Daily rainfall in Fallon during May through October for each year of leaf collection, 2007 through 2010. Short marks indicate days with only a trace of rainfall (<0.25 mm). Data from NCDC, NOAA. Graph for 2007 is modfied from Sheppard et al. (2009a).

Trees were selected for sampling at differing spatial densities from one year to the next. In 2008 and 2009, some trees were sampled within 0.25 km of the hard-metal facility, i.e., very near to it, while others were sampled farther away (Fig. 1b,c). In 2010, trees were sampled along a more even continuum of distance from the hard-metal facility (Fig. 1d). Sampling near the hard-metal facility does not represent biased sampling on our part, as has been suggested by others (Sueker, 2006). Rather, this is an example of targeted sampling (e.g.,

Seinfeld, 1972) to take advantage of prior knowledge that the area of the hard-metal facility is known to be where peak loadings of airborne tungsten and cobalt have occurred and where spatial variability in airborne tungsten and cobalt has been high (Sheppard et al., 2006b). In each year 2008–2010, leaves from just 10 trees were sampled, as opposed to the 95 trees sampled in 2007 (Sheppard et al., 2009a). It is no longer necessary to sample extremely intensively to discern patterns of airborne tungsten and cobalt in Fallon. Geographic coordinates were recorded for each sampled tree to facilitate mapping.

From each tree sampled, an outer branchlet of several leaves was clipped off with pruning cutters from a height of about 2 m above ground. The aspect of each tree sampled was not held constant or patterned, so sampling was effectively random across trees. None of the trees sampled was next to other trees, so there is no forest canopy effect in this study (Dasch, 1987). Branchlets were stored in clean paper bags during fieldwork. Later, leaves were trimmed from their petioles using clean, ceramic (non-metal) scissors.

2.3 Chemical measurement

Leaf tissues themselves were not measured for metals content, but rather rinse water solutions of particulates from the leaf surfaces were measured. Consequently, this study reflects airborne metals that accumulate on leaf surfaces, not soil-derived metals that move through the trees to leaf tissues (Wolterbeek and Bode, 1995). Trimmed leaves were placed in clean, 50-ml polypropylene vials, and tepid, de-ionized water was added to completely submerge the leaves. The vials were capped tightly and shaken lightly for two hours (Little, 1973). Rinse solutions were poured into new, clean polyurethane vials. Rinsed leaves were then oven dried at 50° C for several days and weighed to ±0.0001 g.

Rinse solutions were filtered with acid-washed GHP Acrodisc syringe filters (less than 0.2 μm) and acidified to pH less than 2 with certified pure nitric acid. Measurement was performed using inductively coupled plasma, mass spectroscopy ICP-MS (Elan DRC-II, Perkin Elmer, Shelton, CT). Most analytes were measured in standard mode (e.g., vanadium, nickel, copper, zinc, arsenic, cadmium, cesium, tungsten, lead, and uranium), while chromium and cobalt were measured using the dynamic reaction cell (DRC) flushed with ammonia gas. For all elements measured, detection limits were sub-ppb based on three standard deviations from the mean of 11 replicate measurements.

2.4 Quantitative analysis

Measured contents of metals in rinse solutions were standardized to oven dry mass of leaves measured. Oven dry leaf masses were first transformed to the ¾ power before this standardization step. This transformation incorporates a concept of diminishing returns whereby leaf area increases ever more slowly as leaf mass increases, i.e., at a power of ¾ of leaf mass (Niklas & Cobb, 2008). This transformation was not done originally with the 2007 leaf collection (Sheppard et al., 2009a), but it has since been done on that data set in addition to the data from 2008–2010.

The 2008 and 2009 collections were analyzed by testing medians of element concentrations from trees near the hard-metal facility versus trees away from it. The Mann-Whitney test of medians (Sokal & Rohlf, 1981) was used to assess significance between near trees versus far trees, with the alternative hypothesis being one-tailed, i.e., that element concentrations of trees near the hard-metal facility were higher than those of trees farther away. The 2010 collection was analyzed by plotting concentration values by distance from the hard-metal facility and assessing the strength of a power function model for each element.

3. Results

3.1 Year 2008

From leaves of 2008, tungsten and cobalt show the largest differences between trees near the hard-metal facility versus trees away from it. Tungsten and cobalt medians were 38 times and 16 times more concentrated on surfaces of leaves of trees near the facility than trees far away (Table 1). These differences are highly significant.

element	Concentration ($\mu g \cdot g^{-(3/4)}$) Near (n=6)	Far (n=4)	Near: Far	p value
tungsten	0.793	0.021	37.76	0.007
cobalt	2.052	0.131	15.66	0.007
nickle	0.526	0.040	13.15	0.021
cesium	0.045	0.004	11.25	0.021
sodium	3934	406	9.69	0.035
tantalum	0.0045	0.0005	9.00	0.007
rubidium	7.53	0.94	8.01	0.021
magnesium	2603	329	7.91	0.055
zirconium	0.051	0.011	4.64	0.007
lead	0.037	0.008	4.63	0.013
silicon	331	87	3.80	0.035
beryllium	0.0018	0.0005	3.60	0.143
copper	0.596	0.187	3.19	0.055
phosphorus	631	218	2.89	0.120
titanium	2.18	0.78	2.79	0.083
molybdenum	0.086	0.032	2.69	0.035
vanadium	0.067	0.028	2.39	0.007
strontium	18.0	7.7	2.34	0.169
zinc	3.00	1.32	2.27	0.013
potassium	17229	7715	2.23	0.083
chromium	0.021	0.010	2.10	0.035
gallium	0.036	0.018	2.00	0.228
barium	1.26	0.76	1.66	0.228
lithium	14.9	10.3	1.45	0.457
calcium	5063	3554	1.42	0.169
arsenic	0.188	0.157	1.20	0.375
aluminum	8.7	10.6	0.82	Ø
barium	264	410	0.64	Ø
manganese	37	207	0.18	Ø
selenium	0.001	0.027	0.04	Ø
iron	18	563	0.03	Ø

Table 1. Results for the 2008 collection, sorted by the ratio Near:Far, i.e., the ratio of the median value of trees near the hard-metal facility versus trees farther away from it. Concentration values are medians. P value is for Mann-Whitney testing of no difference between median values versus the one-tailed alternative that the near median is greater than the far median (Sokal & Rohlf, 1981).

Other elements measured show smaller differences between trees near the hard-metal facility versus trees away from it. Most elements show near:far ratios of less than 10, and most of these differences are less significant statistically than the differences for tungsten and cobalt (Table 1). Of these other elements, nickel and cesium show the highest near:far ratios, both above 10. Cesium showed notable spatial variability in the 2007 collection (Sheppard et al., 2009a), and this replication of spatial variability in cesium makes that element potentially interesting environmentally in Fallon.

3.2 Year 2009

From leaves of 2009, tungsten and cobalt show the largest differences between trees near the hard-metal facility versus trees away from it. Tungsten and cobalt medians were 12 times and 7 times more concentrated on surfaces of leaves of trees near the facility than trees far away (Table 2). These differences are highly significant.

Other elements measured show smaller differences between trees near the hard-metal facility versus trees away from it. Other elements show near:far ratios of 3.0 or less, and most of these differences are less significant statistically than the differences for tungsten and cobalt (Table 2). No other element measured shows spatial variability that is especially notable or potentially interesting environmentally in Fallon.

3.3 Year 2010

From leaves of 2010, the negative power model of tungsten concentration as a function of distance from the hard-metal facility is very strong. The R^2 value of the tungsten model is 62% (Fig. 3a), much higher than other elements (Fig. 3b-i). The negative power model of cobalt concentration as a function of distance from the hard-metal facility is weaker but still notable. The R^2 value of the cobalt model is 12%, though that is without an obvious high outlying value from the tree that was nine km away from the facility (Fig. 3b).

element	Concentration ($\mu g \cdot g^{(-3/4)}$)		Near: Far	p value
	Near (n=4)	Far (n=6)		
tungsten	0.176	0.015	11.73	0.007
cobalt	0.757	0.113	6.70	0.007
selenium	0.015	0.005	3.00	0.055
calcium	1329	507	2.62	0.120
iron	1.760	0.924	1.90	0.120
chromium	0.009	0.005	1.80	0.083
potassium	2814	1945	1.45	0.297

Table 2. Results for the 2009 collection, sorted by the ratio Near:Far, i.e., the ratio of the median value of trees near the hard-metal facility versus trees farther away from it. Concentration values are medians. P value is for Mann-Whitney testing of no difference between median values versus the one-tailed alternative that the near median is greater than the far median (Sokal & Rohlf, 1981).

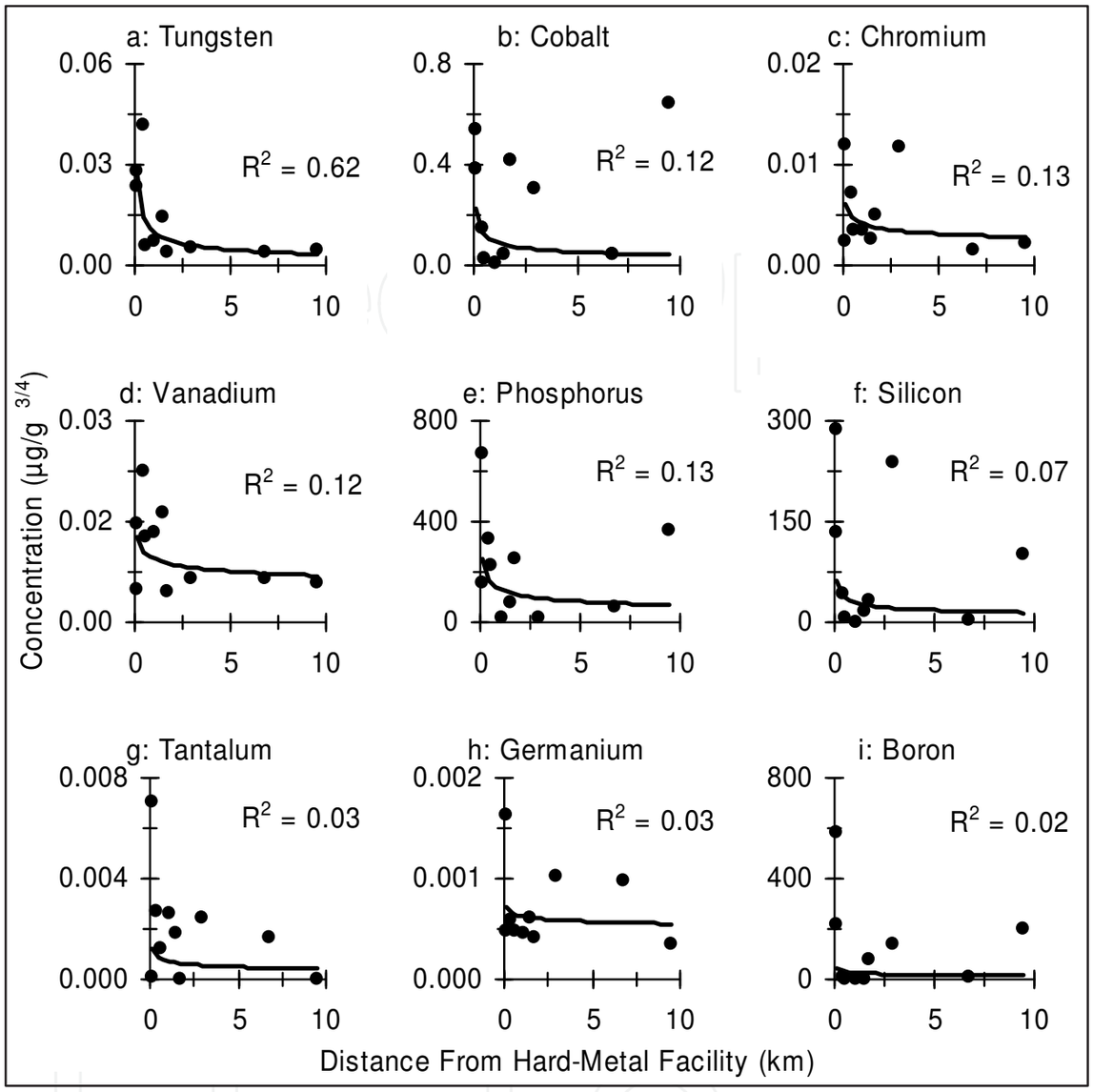

Fig. 3. Leaf surface concentration (mass of element per oven dry mass of leaf raised to the ¾ power) of elements as a function of distance from the hard-metal facility (km) for the 2010 collection. The R^2 value for cobalt is without the outlying value included.

The negative power models of other elements measured are weaker than that of tungsten. Chromium, vanadium, and phosphorus have R^2 values about as strong as that of cobalt (Fig. 3c-e), while other elements have very weak R^2 values (Fig. 3f-i).

3.4 Years 2007–2010

A temporal pattern in absolute maximum values emerged for elements that have been measured all four years. Maximum concentrations of tungsten, cobalt, and chromium have declined fairly steadily from 2007 through 2010 (Table 3). A possible explanation of this trend is that total summer rainfall for Fallon has steadily increased from 2007 through 2010 (Table 3). Maximum concentrations of these three elements show clear negative

relationships with total summer rainfall, each with strong R^2 values (Fig. 4). Rainfall has the obvious potential effect of cleaning leaf surfaces of dust that leaves catch throughout the growing season.

4. Discussion

This work confirms various advantages of leaf-surface chemistry as a technique for assessing air quality. As a relatively easy and inexpensive method, many leaf samples can be collected and analyzed, either for fine-turning spatial variability of air quality at one point in time (Sheppard et al., 2009a) or for assessing temporal variability of air-quality for one area (this study).

Year	Maximum Concentration ($\mu g \cdot g^{-(3/4)}$)			Summer Rainfall (mm)
	Tungsten	Cobalt	Chromium	
2007	16.40	6.70	0.089	8.64
2008	4.78	8.47	0.052	43.43
2009	0.39	1.29	0.021	47.50
2010	0.04	0.64	0.012	64.01

Table 3. Maximum concentration of tungsten, colbalt, and chromium for each year of collecting leaves in Fallon, and total summer (May through October) rainfall recorded at the Fallon Naval Air Station.

Notable findings emerged from this work. One, maximum values of airborne tungsten and cobalt in Fallon have been varying through time at the annual scale. Absolute values from leaf-surface chemistry are standardized for multiple procedural sources of variation (e.g., differing solution volumes and leaf sizes), so the interannual variability in airborne tungsten and cobalt shown here is probably environmental in nature. A logical explanation for this variability is differing summer rainfall totals from year to year.

Two, even during a summer leaf season with high rainfall (i.e., 64 mm in 2010), which logically should reduce absolute values of airborne metals on leaf surfaces, spatial patterns of airborne elements are still discernible with leaf-surface chemistry. This illustrates the robustness of this technique for assessing air quality, at least in environments that are relatively arid to begin with.

In general, long-term environmental monitoring is extremely valuable. Multi-year assessment research allows for discovering slow changes in environmental quality (Johnston, 1991), some of which could have implications for public health. Given the temporal variability of air quality in Fallon, it seems imperative that environmental monitoring and assessment of Fallon, especially for air quality, be continued. Even without considering linkage between exposure to airborne tungsten and cobalt and human illness, merely knowing that airborne tungsten and cobalt are elevated in Fallon and that their airborne loadings are changing through time logically suggests that continuing monitoring and assessment is prudent. Leaf-surface chemistry is a suitable technique for such long-term assessment.

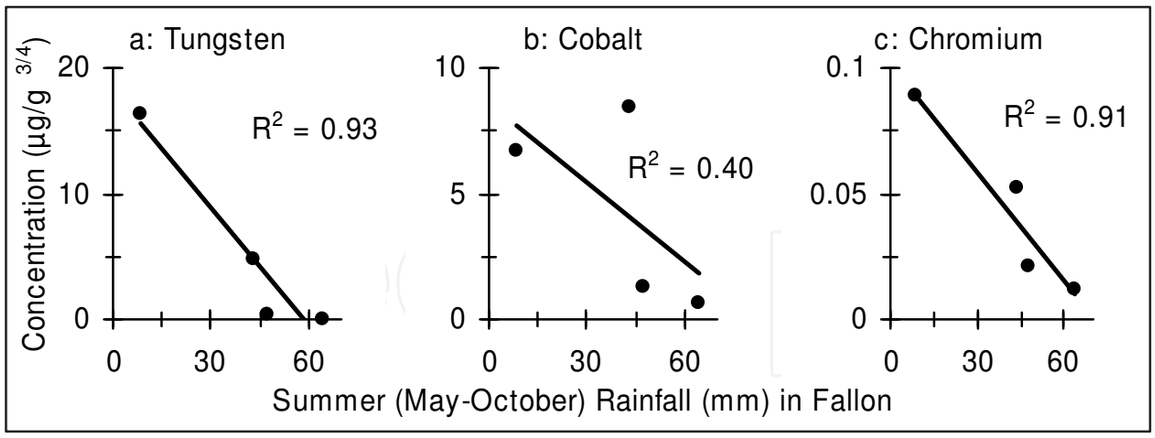

Fig. 4. Maximum concentration (mass of element per oven dry mass of leaf raised to the ¾ power) of tungsten (a), cobalt (b), and chromium (c) as a funtion of total summer (May–October) rainfall (mm) recorded in Fallon for each year (2007–2010).

5. Conclusion

Leaf-surface chemistry is an effective technique for assessing air quality, including for loadings of airborne metals and especially in urban settings. From leaf-surface chemistry, the center area of Fallon is shown to continue having elevated airborne tungsten and cobalt relative to outlying areas around Fallon. Interannual variability in absolute maximum values is notable.

It cannot be concluded from only environmental data that elevated airborne tungsten and/or cobalt cause childhood leukemia. Such a connection requires direct biomedical testing. Nonetheless, given that childhood leukemia in Fallon is the "most unique cluster ever reported" (Steinmaus et al. 2004) and that Fallon is distinctive environmentally by its elevated airborne tungsten and cobalt particulates, it stands to reason that additional biomedical research is warranted to assess the leukogenicity of airborne tungsten and cobalt (e.g., Fastje et al., 2009; Kalinich et al. 2005; Miller et al., 2001; Radcliffe et al., 2010; Steinberg et al. 2007; Sun et al. 2003).

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Court of Nevada, Washoe County, which are related to the childhood leukemia cluster of Fallon. In those cases, the law firm of Dunlap and Laxalt, representing the plaintiffs, with full disclosure to all defendants and their counsel, made an unsolicited donation of $15,000 to assist Witten and Sheppard in furthering their research, with a request that defendants provide similar donations.

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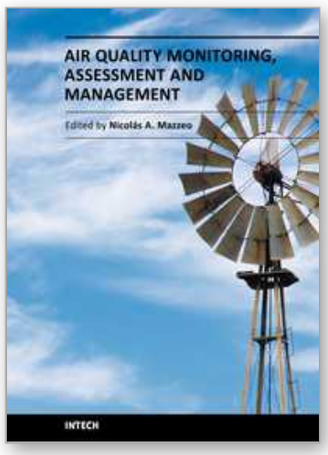

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Human beings need to breathe oxygen diluted in certain quantity of inert gas for living. In the atmosphere, there is a gas mixture of, mainly, oxygen and nitrogen, in appropriate proportions. However, the air also contains other gases, vapours and aerosols that humans incorporate when breathing and whose composition and concentration vary spatially. Some of these are physiologically inert. Air pollution has become a problem of major concern in the last few decades as it has caused negative effects on human health, nature and properties. This book presents the results of research studies carried out by international researchers in seventeen chapters which can be grouped into two main sections: a) air quality monitoring and b) air quality assessment and management, and serves as a source of material for all those involved in the field, whether as a student, scientific researcher, industrialist, consultant, or government agency with responsibility in this area.

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